

IN THE CLAIMS:

1. (Previously presented) A recombinant hybrid virus, comprising:
  - (a) a deleted adenovirus vector genome comprising the adenovirus 5' and 3' *cis*-elements for viral replication and encapsidation; a functional adenovirus genomic region selected from the group consisting of an adenovirus E1a region, E2a region, E4orf6 region, VA RNA region, and any combination of the foregoing; and further comprising a deletion in an adenovirus genomic region selected from the group consisting of:
    - (i) the polymerase region, wherein said deletion essentially prevents the expression of a functional polymerase protein from said deleted region and said hybrid virus does not otherwise express a functional polymerase protein,
    - (ii) the preterminal protein region, wherein said deletion essentially prevents the expression of a functional preterminal protein from said deleted region, and said hybrid virus does not otherwise express a functional preterminal protein, and
    - (iii) both the regions of (i) and (ii); and
  - (b) a recombinant adeno-associated virus (AAV) vector genome flanked by the adenovirus vector genome sequences of (a), said recombinant AAV vector genome comprising (i) AAV 5' and 3' inverted terminal repeats, (ii) an AAV packaging sequence, and (iii) a heterologous nucleic acid sequence, wherein said heterologous nucleic acid sequence is flanked by the 5' and 3' AAV inverted terminal repeats of (i), and further wherein the AAV vector genome does not encode the AAV Rep or AAV capsid proteins,wherein upon infection of a 293 helper cell line, the recombinant hybrid virus is packaged into an AAV particle essentially without producing contaminating adenovirus.

2. (Previously presented) The recombinant hybrid virus of Claim 1, wherein said adenovirus 5' and 3' *cis*-elements comprise 5' and 3' adenovirus inverted terminal repeats and an adenovirus packaging sequence.
3. (Original) The recombinant hybrid virus of Claim 1, wherein said AAV inverted terminal repeats are selected from the group consisting of AAV-1, AAV-2, AAV-3, AAV-4, AAV-5 and AAV-6 inverted terminal repeats.
4. (Canceled)
5. (Original) The recombinant hybrid virus of Claim 1, wherein said adenovirus vector genome comprises sequences encoding an AAV Rep protein.
6. (Original) The recombinant hybrid virus of Claim 5, wherein said sequences encoding said AAV Rep protein are operably associated with an inducible promoter.
7. (Currently amended) The recombinant hybrid virus of Claim 6, wherein said inducible promoter is selected from the group consisting of a tetracycline response element, an ecdysone response element, a heat shock promoter, an MMLV long terminal repeat sequence, a bacteria phage T7 promoter, a metallothionein response element, and the AAV p5 promoter.
8. (Currently amended) The recombinant hybrid virus of Claim 5, wherein said sequences encoding said AAV Rep protein are operably associated with a promoter selected from the group consisting of a liver-specific promoter, a muscle-specific promoter, and a brain-specific promoter.
9. (Original) The recombinant hybrid virus of Claim 1, wherein said adenovirus vector genome comprises sequences encoding an AAV capsid protein.

10-15. (Canceled)

16. (Original) The recombinant hybrid virus of Claim 1, wherein said adenovirus vector genome further comprises a deletion in an adenovirus region selected from the group consisting of the IVa2 region, the 100K region, the E3 region, the E2a region, the E4 region, the L1 region, the L2 region, the L3 region, the L4 region, the L5 region, the intermediate gene IX region, and any combination of the foregoing, wherein said adenovirus vector genome does not otherwise express a gene product associated with the deleted region.
17. (Original) The recombinant hybrid virus of Claim 1, wherein said adenovirus vector genome comprises a deletion in the preterminal protein region.
18. (Original) The recombinant hybrid virus of Claim 17, wherein said deletion comprises a deletion in the preterminal protein region at about nucleotides 9198 to 9630 of the adenovirus serotype 5 genome or a corresponding region of the genome of an adenovirus of another serotype.
19. (Original) The recombinant hybrid virus of Claim 1, wherein said deletion comprises a deletion in the polymerase region.
20. (Previously presented) The recombinant hybrid virus of Claim 19, wherein said deletion in said polymerase region comprises a deletion at about nucleotides 7274 to 7881 of the adenovirus serotype 5 genome or a corresponding region of the genome of an adenovirus of another serotype.
21. (Original) The recombinant hybrid virus of Claim 1, wherein said heterologous nucleic acid sequence is operatively associated with an expression control sequence.

22. (Original) The recombinant hybrid virus of Claim 21, wherein said expression control sequence comprises a promoter.
23. (Currently amended) The recombinant hybrid virus of Claim 22, wherein said promoter is selected from the group consisting of a liver-specific promoter, a muscle-specific promoter, a brain-specific promoter, and a glucose-responsive promoter.
24. (Original) The recombinant hybrid virus of Claim 22, wherein said promoter is an inducible promoter.
25. (Original) The recombinant hybrid virus of Claim 22, wherein said promoter is selected from the group consisting of the CMV promoter, albumin promoter, EF1- $\alpha$  promoter, P $\gamma$ K promoter, MFG promoter, and Rous sarcoma virus promoter.
26. (Original) The recombinant hybrid virus of Claim 1, wherein said heterologous nucleic acid sequence encodes a polypeptide.
27. (Original) The recombinant hybrid virus of Claim 26, wherein said polypeptide is selected from the group consisting of a therapeutic polypeptide, an immunogenic polypeptide, and a reporter polypeptide.
28. (Original) The recombinant hybrid virus of Claim 27, wherein said polypeptide is associated with a lysosomal storage disease.
29. (Original) The recombinant hybrid virus of Claim 28, wherein said polypeptide is selected from the group consisting of  $\beta$ -galactosidase,  $\beta$ -hexosaminidase A,  $\beta$ -hexosaminidase B, GM<sub>2</sub> activator protein, glucocerebrosidase, arylsulfatase A, galactosylceramidase, acid sphingomyelinase, acid ceramidase, acid lipase,  $\alpha$ -L-iduronidase, iduronate sulfatase, heparan N-sulfatase,  $\alpha$ -N-acetylglucosaminidase acetyl-CoA, glucosaminide acetyltransferase, N-

acetylglucosamine-6-sulfatase, arylsulfatase B,  $\beta$ -glucuronidase,  $\alpha$ -mannosidase,  $\beta$ -mannosidase,  $\alpha$ -L-fucosidase, N-aspartyl- $\beta$ -glucosaminidase,  $\alpha$ -neuraminidase, lysosomal protective protein,  $\alpha$ -N-acetyl-galactosaminidase, N-acetylglucosamine-1-phosphotransferase, cystine transport protein, sialic acid transport protein, the CLN3 gene product, palmitoyl-protein thioesterase, saposin A, saposin B, saposin C, and saposin D.

30. (Original) The recombinant hybrid virus of Claim 26, wherein said polypeptide is associated with a glycogen storage disease.
31. (Original) The recombinant hybrid virus of Claim 30, wherein said polypeptide is selected from the group consisting of glucose 6-phosphatase, lysosomal acid  $\alpha$  glucosidase, glycogen debranching enzyme, branching enzyme, muscle phosphorylase, liver phosphorylase, phosphorylase kinase, muscle phosphofructokinase, glycogen synthase, phosphoglucoisomerase, muscle phosphoglycerate kinase, phosphoglycerate mutase, and lactate dehydrogenase.
32. (Original) The recombinant hybrid virus of Claim 31, wherein said polypeptide is a lysosomal acid  $\alpha$ -glucosidase.
33. (Original) The recombinant hybrid virus of Claim 32, wherein said polypeptide is a human lysosomal acid  $\alpha$ -glucosidase.
34. (Original) The recombinant hybrid virus of Claim 1, wherein said heterologous nucleic acid sequence encodes an antisense nucleic acid sequence.
35. (Currently amended) A hybrid virus particle comprising the recombinant hybrid virus of Claim 1 encapsidated within an ~~adeno-virus~~ adenovirus capsid.
36. (Previously presented) A cell comprising the recombinant hybrid virus of Claim 1 or the hybrid virus particle of Claim 35.

37. (Original) A method of producing a recombinant adeno-associated virus (AAV) particle, comprising providing to a cell:
- (a) a recombinant hybrid virus according to Claim 1 or a hybrid virus particle according to Claim 35;
  - (b) AAV sequences sufficient for replication and packaging of the AAV vector genome; and
  - (c) AAV sequences sufficient to produce a functional AAV capsid, wherein (a), (b) and (c) are provided to the cell under conditions sufficient for replication of the AAV vector genome and packaging thereof in the AAV capsid such that AAV particles comprising the AAV vector genome encapsidated within the AAV capsid are produced in the cell.
38. (Original) The method of Claim 37, further comprising the step of collecting the recombinant AAV particle.
39. (Original) The method of Claim 37, further comprising providing to the cell the adenovirus helper functions for AAV replication and packaging.
40. (Original) The method of Claim 39, wherein adenovirus E1a, E2a, E4orf6, and VA RNA helper sequences are provided.
41. (Original) The method of Claim 37, wherein the cell is selected from the group consisting of a HeLa cell, a 293 cell, a muscle cell, and a liver cell.
42. (Original) The method of Claim 37, wherein essentially no adenovirus particles are produced.
43. (Original) The method of Claim 37, wherein the yield of recombinant AAV particles is at least 5-fold greater than in the presence of the adenovirus polymerase and/or preterminal proteins.

44. (Original) The method of Claim 37, wherein sequences encoding an AAV Rep protein and/or sequences encoding the AAV capsid protein are stably expressed by the cell.
45. (Original) The method of Claim 37, wherein sequences encoding an AAV Rep protein and/or sequences encoding the AAV capsid protein are provided by a vector other than the recombinant hybrid virus.
46. (Original) The method of Claim 45, wherein the vector is selected from the group consisting of a plasmid, an adenovirus, an Epstein Barr virus, and a herpesvirus vector.
47. (Original) The method of Claim 37, wherein the AAV inverted terminal repeats and the AAV capsid are derived from different AAV serotypes.
48. (Original) The method of Claim 37, wherein the AAV capsid is an AAV-6 capsid.
49. (Original) The method of Claim 37 or Claim 48, wherein the AAV inverted terminal repeats are AAV-2 inverted terminal repeats.
50. (Original) A method of producing a recombinant adeno-associated virus (AAV) particle, comprising providing to a cell a hybrid virus particle according to Claim 35, said recombinant hybrid virus particle expressing the adenovirus helper functions for AAV replication and packaging; wherein the cell (i) expresses AAV *rep* sequences sufficient for replication and packaging of the AAV vector genome, (ii) expresses AAV *cap* sequences sufficient to produce a functional AAV capsid, and (iii) does not express sequences sufficient to produce a functional adenovirus E1a protein; and further wherein the hybrid virus particle is provided under conditions sufficient for replication of the AAV vector genome and packaging thereof in the AAV capsid such that AAV particles comprising the AAV vector genome encapsidated within the AAV capsid are produced in the cell.

51. (Original) A method of producing a recombinant adeno-associated virus (AAV) particle, comprising providing to a cell a hybrid virus particle according to Claim 35, the hybrid virus particle expressing:

- (i) adenovirus helper functions for AAV replication and packaging except the hybrid virus particle does not express a functional adenovirus E1a gene product,
- (ii) AAV *rep* sequences sufficient for replication and packaging of the AAV vector genome, and
- (iii) AAV *cap* sequences sufficient to produce a functional AAV capsid,

wherein the cell expresses functional adenovirus E1a gene products; and further wherein the hybrid virus particle is provided to the cell under conditions sufficient for replication of the AAV vector genome and packaging thereof in the AAV capsid such that AAV particles comprising the AAV vector genome encapsidated within the AAV capsid are produced in the cell.

52. (Original) A method of producing a recombinant adeno-associated virus (AAV) particle, comprising providing to a cell:

- (a) a hybrid virus particle according to Claim 35, the hybrid virus particle expressing adenovirus helper functions for AAV replication and packaging except the hybrid virus particle does not express a functional adenovirus E1a gene product,
- (b) a separate vector comprising inducible AAV *rep* sequences sufficient for replication and packaging of the AAV vector genome, and AAV *cap* sequences sufficient to produce a functional AAV capsid,

wherein the cell expresses a functional adenovirus E1a gene product; and further wherein (a) and (b) are provided to the cell under conditions sufficient for replication of the AAV vector genome and packaging thereof in the AAV capsid such that AAV particles comprising the AAV vector genome encapsidated within the AAV capsid are produced in the cell.



53. (Original) A method of producing a recombinant adeno-associated virus (AAV) particle, comprising providing to a cell:
- (a) a hybrid virus particle according to Claim 35, the hybrid virus particle expressing adenovirus helper functions for AAV replication and packaging,
  - (b) a separate vector comprising AAV *rep* sequences sufficient for replication and packaging of the AAV vector genome, and AAV *cap* sequences sufficient to produce a functional AAV capsid,
- wherein the cell does not express a functional adenovirus E1a gene product; and further wherein (a) and (b) are provided to the cell under conditions sufficient for replication of the AAV vector genome and packaging thereof in the AAV capsid such that AAV particles comprising the AAV vector genome encapsidated within the AAV capsid are produced in the cell.
54. (Original) The method of Claim 52 or Claim 53, wherein the separate vector is a plasmid vector.
55. (Original) The method of Claim 52 or Claim 53, wherein the separate vector is an adenovirus vector.
56. (Original) A method of introducing a nucleic acid into a cell, comprising contacting a cell with the recombinant hybrid virus of Claim 1 or the hybrid virus particle of Claim 35 under conditions sufficient for entry of the recombinant virus particle into the cell.
57. (Original) The method of Claim 56, wherein the cell is selected from the group consisting of a neuron, a brain cell, a retinal cell, an epithelial cell, a cardiac muscle cell, a smooth muscle cell, a skeletal muscle cell, a diaphragm muscle cell, a pancreatic cell, a liver cell, a fibroblast, an endothelial cell, a germ cell, a lung cell, a prostate cell, a stem cell, a progenitor cell, and a cancer cell.
58. (Original) The method of Claim 56, wherein the cell is a mammalian cell.

59-66. (Canceled)

67. (Previously presented) The method of Claim 56, further comprising introducing an AAV Rep 68/78 protein or sequences encoding an AAV Rep 68/78 protein into the cell.

68-142. (Canceled)

143. (Original) The recombinant hybrid virus of claim 1, wherein the deleted adenovirus vector genome further comprises a functional adenovirus E1a region.

144. (Original) A recombinant cell comprising the recombinant hybrid virus of claim 1, wherein the recombinant cell is stably modified to express a functional polypeptide, a functional pTP polypeptide, or both.

145. (Currently amended) A recombinant hybrid virus, comprising:

- (a) a deleted adenovirus vector genome comprising:
  - (i) the adenovirus 5' and 3' *cis*-elements for viral replication and encapsidation;
  - (ii) a functional E1a region and at least one additional functional adenovirus genomic region selected from the group consisting of an adenovirus E2a region, an adenovirus E4orf6 region, and an adenovirus VA RNA region; and
  - (iii) a deletion in an adenovirus genomic region, wherein the deletion essentially prevents the expression of a functional polymerase protein, a functional preterminal protein, or both from the adenovirus genomic region, and further wherein said hybrid virus does not otherwise express a functional polymerase protein, a functional preterminal protein, or both; and

- (b) a recombinant adeno-associated virus (AAV) vector genome flanked by the adenovirus vector genome sequences of (a), said recombinant AAV vector genome comprising:
    - (iv) a heterologous nucleic acid sequence flanked by AAV 5' and 3' inverted terminal repeats;
    - (v) an AAV packaging sequence; and
    - (vi) a deletion of the AAV Rep region, the AAV Cap region, or both, such that the ~~the~~ recombinant AAV vector genome does not encode a functional AAV Rep polypeptide, a functional AAV capsid polypeptide, or both,wherein upon infection of a 293 helper cell line, the recombinant hybrid virus is packaged into an AAV particle essentially without producing contaminating adenovirus.
146. (Previously presented) A hybrid virus particle comprising the recombinant hybrid virus of claim 145 encapsidated within an adenovirus capsid.
147. (Previously presented) An adeno-associated virus (AAV) particle produced by introducing the hybrid virus particle of claim 146 into a cell that expresses AAV Rep and AAV Cap.
148. (Original) A recombinant cell comprising the recombinant hybrid virus of claim 145, wherein the recombinant cell is stably modified to express a functional polypeptide, a functional pTP polypeptide, or both.